IOSR Journal of Environmental Science, Toxicology and Food Technology (IOSR-JESTFT)
e-ISSN: 2319-2402,p- ISSN: 2319-2399.
Volume. 1 Issue 6, PP 45-49
www.iosrjournals.org

To Study the Diversity of Fungal Species in Sewage Water of Durg District

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Abstract: *The present study aimed to find out the fungal diversity of sewage water of some selected area in Durg city. Before isolation of fungi the sewage water samples were analyzed for different physico- chemical characteristics.To isolate the fungi in different sewage water samples, sterilization technique, serial dilution and spread plate technique were used.Presence of fungi was detected in the different samples on the basis of morphological characteristics and colony forming units. The results obtained showed that most widely distributed fungi in sewage water samples were Aspergillus sp.*

Keywords: *Fungal diversity, Isolation, Physico-chemical, Sewage*

I. Introduction

Water is one of the naturally occurring essential requirements of all life functions. It is a master solvent, and all metabolic reactions of living beings depend mainly on its presence. As we know, human population living in towns, cities etc., depends upon municipal supplies of water. Besides, a major fraction of our population which lives in rural areas, especially in underdeveloped and developing countries, depends upon lakes, rivers, ponds, springs, wells etc. for their water requirements. But today water pollution has become one of the major problems for all life forms.Water pollution is an appalling problem, powerful enough to lead the world on a path of destruction. Water is an easy solvent, enabling most pollutants to dissolve in it easily and contaminate it.Contamination of water starts right from the beginning when water reaches the earth through air in the form of precipitation; microorganisms present in air get entry into it. After the precipitation is over and water reaches the earth surface, it gets contaminated by microorganisms via soil, dead plants and animals etc. In addition, the natural water supply sources are getting contaminated by a large array of substances such as domestic and industrial wastes i.e., sewage discharged as a consequence of civilized man's need, and human and animal excreta in the form of urine and faces.It is important to note that these contaminations, particularly domestic and industrial waste and fecal ones, are of much microbiological concern because they not only increase the biochemical oxygen demand (BOD) but also sometimes contain certain disease-causing microorganisms.

Durg is a major city in Chhattisgarh state, central India east of the Shivnath River and is part of the Durg-Bhilai urban agglomeration. The city is an agricultural market and heavily engaged in milling rice. Durg gained importance as an industrial centre after the establishment of a large steel plant at Bhilai. Other industries include brass working and bell-metal working, oil pressing, mining and weaving. It is the third largest district of Chhattisgarh. As it is one of the developing citiesof the state and has many industries and known for large urban area, there is a large disposal of industrial and household waste effluent leading to raise the sewage water.

Sewage is a liquid or solid wastes carried off in sewers. It consists of domestic water-borne wastes including human and animal excreta, washing waters and everything that goes down the drains of a town or a city. It also consists of industrial water-borne wastes as well as ground, surface and atmospheric waters which enter the sewerage system.The microbial population per milliliter of sewage may vary from a few lacs to several millions. Various types of microorganisms, viz., microfungi, bacteria and protozoa, collectively called 'sewage fungus' are known to grow profusely in sewage. In present study an attempt is made to determine some physiochemical properties sewage water and to know the diversity of fungi in it.

II. Material and method

and Mohan Nagar(S5) of Durg cityand were analyzed for physico-chemical characteristics by the methods of WHO and APHA AWWA (1985).Sewagewater samples were taken in plastic bottles separately.For isolation of fungi sterilization technique, serial dilution and spread plate techniques were used.

1.1. Physio-chemical analysis

The sewage water samples were analyzed for physico-chemical properties taking different parameters. These parameters are temperature, pH, acidity, alkalinity, hardness, phenol, DO, COD, BOD and MPN

1.2. Sterilization technique

Petri plates, conical flasks, test-tubes and other glassware were sterilized in autoclave. For sterilization purpose all apparatus were autoclaved for 30 minutes at 121°C. After autoclaving all sterilized material was dried in an oven at 90°C.

1.3. Media preparation

Potato Dextrose Agar (PDA) media was used for fungal cultures growth (Razak et al., 1999). Two hundred grams of potato were peeled, sliced, boiled and then sieved through a clean Muslin cloth to get a broth in which agar and glucose were added. The media was then autoclaved for 30 minutes at 121°C. To suppress the bacterial growth 0.5 ml/L streptomycin was added in the medium (Martin, 1950).

1.4. Dilution preparation

The purpose of serial dilution was to the colonies of fungi. One ml of sewage water wastaken from each sample. Serial dilution was set up by carefullytaking the 10 ml of distilled water in McCartney bottles. Thenthese bottles were autoclaved for 30 minutes at 121°C. From the sample of sewage water 1 ml was dissolved in 10 ml of steriledistilled water in McCartney bottle to give (1:10) and shakedwell. The McCartney Bottle 2 was inoculated with 1 ml frombottle 1 to give 1:100 dilutions. McCartney Bottle 2 was alsoshaked well. McCartney Bottle 3 was inoculated with 1 ml frombottle 2 to give 1:1000 dilutions. McCartney Bottle 4 wasinoculated with 1 ml from bottle 3 to give 1:10000 dilutions.To complete the serial dilutionmicropipette was used with sterilized tips. Estimation of fungalpopulation was done by standard spread plate dilution methoddescribed by Seeley and Van Denmark (1981) in triplicates.

1.5. Isolation of fungi

Spread plate technique was used for enumeration of fungifrom given samples. From each McCartney bottle 0.5 ml ofsample was taken separately with the help of micropipette along with sterilized tips. Then these diluted samples wereinoculated on sterile PDA plates with the help of micropipetteand L shape rod was used to spread the diluted sample on the PDA plate. The same step was repeated with all other sewage water samples. Then these plates were incubated at 30°C for 3 days and then the colonies were counted (Adesemoye et al.,2006).

1.6. Identification of fungi

The cultures were identified at genus level on the basis ofmacroscopic (colonial morphology, color, texture, shape andappearance of morphology) and microscopic characteristics(septation in mycelium, presence of specific reproductive tructures, shape and structure of conidia) (Zafar et al., 2006).

III. Results and Discussion

1.7. Physico-chemical analysis

The physico-chemical quality of sewage water totally depends on the geological condition of the soil and ground water pollution of the area. The physico-chemical parameters ofsewage water of different sites inDurg district are shown in Table -1 which shows a vast range in properties of samples from different sites.

1.8. Morphological characteristics of fungi

From the collected sewage water samples three major speciesof*Aspergillus* and other eight species were isolated. Table 2 shows the morphologicalcharacteristics and this indicates that *Aspergillusniger*istypically black and different from the other species of the*Aspergillus.Aspergillusflavus*is yellow green while *Aspergillusfumigatus* is green. While other five species*Curvularia,Chaetomium,Acremonium, Ttrichoderma*and*Fusarium* were different in color and morphological characteristics.

1.9. Occurrence of number of colonies of fungi

From the collected samples, occurrence of species indifferent samples is shown in Table 3, which shows the total number of colonies of different fungal species obtained in sewage water samples of different sites.

A total of 233 colonies of fungal species were recorded from sewage water samples of five different sites. It can be clearly observed that among 8 fungal species the highest number of colony obtained is of *Aspergilusniger*. Total 64 colonies of *Aspergilusniger* were recorded. This shows that in all samples the presence of *Aspergilusniger* is very frequent. The second frequent species found is *Aspergillusflavus*, total 35 colonies of it were recorded. The total number of colonies of *Aspergillusfumigates* obtained was 31.Total 29 colonies of *Acremonium sp.* was recorded.The total number of colonies of *Chaetomium sp*.obtained was 28.The total number of colonies of both *Trichoderma sp.* and *Fusariumsp.*obtained was 23 respectively.

IV. Conclusion

From the results it was concluded that different sewage water has large variation in their physico-chemical characteristics which is due tothe environmental condition, geographical area of that region and soil texture of the area. The present study has built initial knowledge on fungal diversity in sewage water. On the basis of number of fungal colonies obtained during the study it can be stated that sewage water contains a vast number of different fungi.

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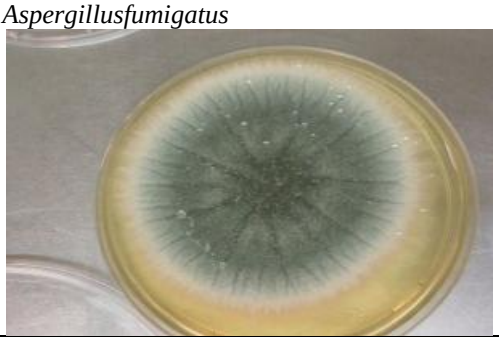
Table 1. Physico-chemical analysis of sewage water samples of five different sites

S. NO	Parameters	Borsi (S1)	Padmanabhpur(S2)	Subha snagar (S3)	Potiya (S4)	Mohan nagar (S5)
1	Temperature	33◦	31◦C	33◦C	30◦C	32◦C
2	Ph	7.3	7.4	7.6	7.3	7.4
3	Acidity	24.59	26.46	34.26	28.6	25.76
4	Alkalinity	36.15	16.72	10.84	28.4	64.47
5	Hardness	401.2	206.8	302.4	350.4	781.6
	Ca	329.2	156	204	941.2	626.4
	Mg	72	50.8	98.4	-590.8	155.2
6	Phenol	0.5	0.1	0.2	0.4	0.1
7	DO	6.4	4.4	0.4	4	4.4
8	COD	153.6	280	232	144	208
9	BOD	0.03	0.03	1.97	0.07	0.03
10	MPN	2	2	7	20	7

Table 2. Morphological characteristic of isolated fungi from the sewage water

Species	Morphological Characteristic
Aspergillusniger 	Typically black powdery colony, large conidia.
Aspergillusflavus 	Yellow green to brown colony, conidiophores hyaline.
Aspergillusfumigatus 	Green colony, columnar conidial heads, pigmentedConidiophores.
Chaetomiumsp 	Colony color is from white, grey to red and brown, hyphaeareseptate.
Acremoniumsp	White, grey, pink, rose or orange in color, hyphae are fineand hyaline.

	
Trichodermasp  .	Mostly white color colony, conidiophore are yellow in colorhighly branched loosely or compactly tufted.
Fusariumsp 	White, red and pink colour colony, septate hyphae.

Table 3.No. of isolated colonies of fungi

Fungal Sp.	S1	S2	S3	S4	S5	Total No. of colonies
Aspergillusniger	18	10	9	12	15	64
Aspergillusflavus	3	7	10	8	7	35
Aspergillusfumigatus	5	3	8	10	5	31
Chaetomium sp.	9	5	3	7	4	28
Acremonium sp.	6	8	5	3	7	29
Trichoderma sp.	3	5	7	4	4	23
Fusarium sp.	6	3	5	4	5	23