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ORIGINAL ARTICLE

Analysis of Microorganisms in Chambal River at Dholpur District

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ABSTRACT

Biomonitoring is a reliable, convenient and quick method for assessing the water quality. Relative bacterial concentration as well as Zooplankton and phytoplankton serve as good indicator of water quality. Concentration of E-coli graph in water is a valid evidence for determining the load of feacal matter. The samples of water may be collected from time to time from clean polluted sites respectively as per described method for collection of water samples for plankton and bacterial analysis. Thus it is obvious after determining the physico-chemical quality the analysis for different types of microorganisms may be done and tabulated in the present study.

***Key words**: Bacteria, Fungus, Protozoa, Chambal River*

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INTRODUCTION

Climate change is a critical environmental issue of our time. Carbon dioxide is a greenhouse gas, and the new view of freshwaters as hotspots for carbon transformations has taught scientists more about how freshwater systems, such as rivers, may fit into the global carbon budget. Dissolved organic matter (DOM) is a critical intermediate in the global carbon cycle because it is the largest pool of reduced organic matter transferred from land to water that has the potential to be oxidized by bacteria and sunlight to CO2. Researchers at the UNC Department of Marine Science have conducted studies right here in North Carolina on the Neuse, Tar-Pamlico, Roanoke and Haw Rivers. They examine the source, quantity and availability of organic matter and the extent to which this organic matter is processed by freshwater microbial communities. Researchers at UNC Environmental Science and Engineering, Gillings School of Global Public Health, are studying the release of CO2 from thawing soils in the Arctic. They examine the photochemical and biochemical interactions that affect DOM and, in turn, the transformation of DOM to atmospheric CO2. Recent studies show that the release of carbon dioxide into the atmosphere from freshwaters is more important in the global carbon cycle than previously recognized. The conventional model was that stream and rivers acted as "pipes" passively conveying terrestrial carbon (produced by decomposing plants) to the ocean. The new view is that freshwaters are "hotspots" for the transformation of terrestrial carbon while it makes its long journey from headwater streams to the ocean. Scientists now know that at least 100 million metric tons of CO2 per year is released into the atmosphere from rivers and streams during terrestrial carbon's journey to the oceans. By comparison, motor vehicles currently emit over 900 million metric tons of CO2 worldwide each year.

MATERIALS AND METHODS

SAMPLE FOR BACTERIAL ANALYSIS:

For bacteriological analysis of collection of samples required more precautions. Generally samples are collected from the marginal waters such type of water is disturbed due to contaimination by various human activities. There is a marked variation in total bacterial density, coliform population and the density of entrobacteria in marginal waters and in water away from the margin. For illustrating this point we calculated samples from marginal water and about 100 metre away from the margin between October to July.

Table 1: The average reading

Parameter	100 metre away	Margin
Total coliform/100 ml	4,600	16,000
Total bacterial density Density X10^7/litre	0.171	0.687
Density of Entrobacteria 10^5/litre	0.13	1.5

The above readings clearly indicate show about 4 times increase in bacterial density and coliform number in marginal waters. Collected samples are put in ice box immediately and recorded. It is lesser therefore cleared that water sample for bacterial analysis should not be collected from near the bathing ghat, Ferry stands or other, distributed point so the sample have collected from B, C and D points for bacterial analysis. We take narrow mouthed glass tubes of 250 ml capacity (plate) and collected water samples in it then sodium thiosulphate solution should be pipetted in these glass tubes for dechlorination.
The mouth of these tubes should be wrapped by thick paper. The bottles along with the solution be sterilized in an autoclave at 15-16 pressure for 15 minutes. At the time of sample collection the base of the bottle should be immersed in water in oblique angle and in an anticurrent direction. The stopper should be removed and the water sample should be immediately collected. After collection the stopper should be tightly fitted and it should be covered by small plastic shut and keep it in the ice box. This sample should be brought to the laboratory at the shortest possible time. In case the ice starts melting, more ice cubes should be added in ice box.

TOTAL BACTERIAL DENSITY

This is done by serial dilution plate count method. In this method dilute the sample, 1.0 ml of water is diluted in stepwise. Sterile quarter strength ringer solution (Sirockin and culliimory 1969) is used as a diluted. If 1.0 ml sample is added in 9.0 ml of sterile ringer's solution the dilution will be 1:10, similarly further dilution may in 1: 100 or 1:100 ratio. The tubes of different dilution samples are thoroughly shaked, subsequently 1 ml of diluted sample from different tubes is taken out with a sterile pipette and keep it in sterile Petri dish. The different samples in different Petri dishes are marked subsequently method, collected and sterilized nutrient agar. Then samples get mixed with Agar. After colling the Petri dishes are incubated in inverted position at 37^0C for 24 to 48 hours. On the Agar plate colonius of bacterias developed which counted with the help of qubee colony counter. In this process the number of bacterias/ml of the original samples multiplied on the average number of colonies by the dilution factor.

FUNGI

Presence of extraneous organic and inorganic materials is a permanent feature of aquatic ecosystem and this leads to major changes in physico chemical and biological components of water. Influx of all ochthonous organic materials through drains, channels due to floods considerably increases the population of the decomposers, specially the bacteria and fungi. The association of bacteria is more common with animal tissues where as the plant tissues are generally colonized by fungal organism due to the rich cellulose and lignin components. Till recently it was considered that flagellate fungi play a dominant role as

decomposers in the aquatic ecosystem, however, now it is established that the dominant role in decomposition is played by the extra aquatic non motile fungi which join the system along with the dead twings, leaves and different part of plants. These fungal organisms ultimately become a part of sediment along with their substrates. Some of the fungi which are frequently encountered in Chambal system are as follows:

HYPOMYCETES

Such extra aquatic fungi contribute to more than 90% of the total fungal population. These fungi play effective role like decomposes in the Chambal ecosystem.Since the Chambal system is very extensive a large number of the air borne fungal conidia may also fall on water surface. Precaution is therefore needed in distinguishing the fungal decomposer from air born fungi. The following procedure is suggested for isolation of fungi associated with decaying and submerged leaves, stem, twigs, grass fragments and seeds etc. These are collected by ordinary cloth net from the surface of water. These fragments should be dried in blotting paper, surface sterilized and then put in culture tubes having PDA medium. The sample of benthic soil may be collected from marginal water (fig) the soil, collected from different locations should be packed in polythene bags. It is different dilutions should be made in lab and plated in petriplate having PDA/C Zapek's medium inoculated petriplates may be placed in sterilized glass chamber's at room temperature for two-three days subsequently the dove love colonies may be isolated for identification of fungi.For isolation of aquatic, hempseed, maize seed, housefly fish pieces etc. are used as baits and this method is known as Bait method. The sample is collected in sterilized Pyrex bottles and then poured in to sterilized petridishes which is then baited with boiled hemp seed, maize seed, housefly or any other suitable baits and incubated at room temperature for 24 hours. After 24 hours the colonized baits are washed thoroughly with sterile distilled water and transferred to fresh sterile distilled water in conical flasks and incubated for 4-7 days. Fungi growing on the baits are isolated preferably in 2% malt extract Agar medium and identified. Regular isolation of fungi by the above two methods and sources will give idea about periodicity of aquatic fungi in the Chambal ecosystem. Separate experiments may be setup in the lab in order to evaluate their role as decomposers of cellulose, lignin and other plant parts. Subsequently, the sample of plankton may be used for evaluating the species diversity index.

SPECIES DIVERSITY INDEX

Species diversity index (H) is computed (Shannon & weaver, 1963) with the help of following: formula

$$H = -E \log \left(\frac{ni}{N} \right)$$

Where,

ni = number of individual of each species

N = total number of individuals

Variation in the population of the living organisms, as a measure of water quality is based on the principal that in clean water community diversity is high while in the polluted waters diversity is low. Wilhm and Dorris (1966) have proposed a relationship between species diversity and polluted status of the sampling sites and defined the Zones as follows:-

Species diversity	Condition
73.0	Clean water
1.0-3.0	Moderately polluted
21.0	Heavy polluted

RESULTS AND DISCUSSION

Table 2: Sitewise arrangement of Bacteria

Bacteria	Site A	Site B	Site C
Micrococcus	-	A	A
Leptothrix	R	D	D
Spirillum	D	D	D
Vibirio	A	D	D
Achromatium	D	D	D
Thiothrix	F	D	A
Myxobacteria	D	-	A
Spirochaeta	F	F	-
Sarcina	F	-	R
Sphaerotilus	R	-	R
Ceptothrix	R	-	-
Chlorobium	R	-	F

D – Dominant, A – Abundant, F – Fluent, R – Rare

Table 3: Sitewise arrangement of Fungi

Fungi	Site A	Site B	Site C
Leptolegina	-	R	R
Loramyces	A	A	A
Lemonniera	R	-	-
Tetrachaetum	-	R	F
Rhizodiomyces	-	-	R
Leptomitirs	F	D	D
Alatospora	R	-	R
Ceriospora	F	F	-
Piricularia	R	F	F
Tricladium	-	R	F
Heliscus	A	R	R
Articulospora	-	R	F

D – Dominant, A – Abundant, F – Fluent, R – Rare

ALGAL POLLUTION INDEX

Besides species diversity the Algae can be used for determining pollution as per method described by Palaner (1969). A score of 20 and above of pollution index was taken for high organic pollution, whereas a score of 15-19 represented a probable evidence of high organic pollution. Lower scores indicated low pollutional level.

Table 4: physicochemical and biological features of polluted and clean water

Test	Polluted water	Clean water
Physicochemical examination	Heavy concentration of nutrients slightly acidic, pH, low D. O., high B.O.D and C.O.D.	Normal nutrients neutral or slightly alkaline pH high D.O., low B.O.D. and C.O.D.
Microscopic examination phytoplankton	Heavy concentration of oscillatoria-chlorine, O. limes, O.tenuis microcystis-aeruginose, choleralla-vulgaris, Ankistrodesmus-falcatus, scendesmus-quadricauda synedra-ulna, Euglena.acus E. viridis, Phacuscaudatus.	Oscillatoria subberevis, Merismopedia-glauca, M. Punctata,Phormidium, Calcacola, Hydrodictyon-reticulatum P. tetras, Cholorococcum sp., Cladophora sp. Synedra-ulna, Euglena.acus E. viridis, Phacuscaudatus
Zooplankton	P. caudatum, Keratella cochleatis, Brachionus plicatilis, B. quadridentata, B. rubens, B. calyciflrus Filinia sps. Platiyas-polyaconthus, Maina-branchiati	
Visual observation	Light pink colonies of moina branchiate (specially in coprogenous environment)	This species is usually not encountered.

ORGANISMS WITH HIGH FREQUENCY IN POLLUTED WATER

Some organisms invariably show a preference for polluted water. These can be used for preparing suitable season wise models and on that basis a relationship can be drown between physicochemical characters of water, bacterial concentration and plankton density/diversity we have found that during summer months the main physicochemical and biological features of polluted and clean water shows in table 4.

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