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<u>Research Article</u>

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ISOLATION AND IDENTIFICATION OF BACTERIA FROM DIFFERENT WATER SAMPLES OF BANKUR DISTRICT OF WEST BENGAL

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ABSTRACT

Water has curious and unusual properties, and plays an important role in living systems. Thus, "no life without water" is common saying. It is a master solvent and all metabolic reaction of the living organism depends on the presence of water. Water gets contaminated with pathogenic micro-organisms through intestinal discharges of man and animal. The characteristics group of intestinal bacteria are the coliforms are defines as facultative anaerobic Gram-negative, non-sporing, rod-shaped bacteria that ferment lactose with gas formation within 48 hours at 35°c, coli forms are the members of the family *Enterobacteriaceae* which includes *E.coli, Enterobacter aerogenes*,

Salmonella and Klebsiella pneumoniae. During present investigation different water sample of Bankura district were collected of West Bengal state. Present work has been carried out to screen the Bacteria of water bodies of Bankura district and to study the characteristics of micro-organisms causing water pollution with regards to their staining behaviour, colony characteristic and biochemical test. The Bankura district lies in the western plateau and high lands. Two sites have been selected for study of micro-organisms and physicochemical characteristics of water. After the continued study from August 09 to April 10, altogether six strains of bacteria were observed and isolated from different water Samples, obtained during present investigation. eg:- *E.coli, Staphylococcus, Streptococcus, Psuedomonas, Shigella,* and *Salmonella typhi* Spp. etc. Some fungi species are also isolated from Water samples eg; *Aspergillus* spp.

KEYWORDS: Bankura, West Bengal, Water, Salmonella, Bacteria, Enterobacter etc.

INTRODUCTION

In aquatic ecosystems, water is an important abiotic component that regulates the formation of assemblage of several interacting organisms, including microbes. Microorganisms are ubiquitous in nature and influence all known ecosystems on earth (Atlas and Bertha, 1998). The ubiquity of microorganisms is attributed mainly to their small size, easy dispersal, adaptation to diverse habitats and ability to utilize wide variety of substrates as nutrient source (Pandey et al., 2007). The members and kinds of micro organisms present in soil depend on many environmental factors such as the amount and type of nutrients, moisture, degree of aeration, pH and temperature etc Sharma et al. (2016). The soil-dwelling microbes can be referred as the "Biological engine" of the earth as they play a pivotal role in many fundamental nutrient cycling processes, soil structure dynamics, pollution degradation and regulation of different plant communities (Breure, 2004). Microbes are also responsible for soil aeration and soil fertility which are among the crucial aspects of soil function. Soil microbial community mainly consists of five major groups, i.e. bacteria, actinomycetes, fungi, algae and protozoa (Holt 1986). Among them bacterial population is generally much higher than other groups (Alexander 1978). To understand the complexity of the interaction mediated by soil microbes, the evaluation of soil microbial diversity is essential.

Water has curious and unusual properties, and plays an important role in living systems. Thus, "no life without water" is common saying. It is a master solvent and all metabolic reaction of the living organism depends on the presence of water (Saha et al., 1971). Nearly three or fourths of the earth's surface is covered by the water mainly oceans to a lesser degree by rivers, lakes, and streams (Chona, 1991). This water is in continuous circulation and the process is known as the water cycle or hydrological cycle. Water is lost from the earth by the way of evaporation, transpiration, exhalation, and is returned to the earth by the way of precipitation (Verma, 2004).

Water gets contaminated with pathogenic micro-organisms through intestinal discharges of man and animal. The characteristics group of intestinal bacteria are the coli-forms are defines as facultative anaerobic Gram-negative, non-sporing, rod-shaped bacteria that ferment lactose with gas formation within 48 hours at 35°c, coli forms are the members of the family *Enterobacteriaceae* which includes *E.coli*, *Enterobacter aerogenes*, Salmonella and *Klebsiella pneumoniae*. *E. coli* is commonly found in the intestinal tract of man and animals, while *E.aerogenes* is normally found on plants and grains and may sometimes occur in the

intestinal tract of man and animals. When such "foreign" enteric indicator bacteria are not detectable in a specific volume (100ml) of water, the water is considered portable water (Latin potadilis- fit to drink) or suitable for human consumption (Potter, 1960). The isolation of specific micro-organisms, which may be present in water, it may involved enrichment media, which encourage growth of the pathogen, while representing the growth of a companying flora, followed by isolation on selective diagnostic media and finally the application of confirmatory test and bases of their biochemical properties, which are compare to the pathogenic micro-organisms.

The studies of the physical-chemical characteristics of Bankura pond are scanty. When looking to the other parts of India, many researchers have already taken this problem. Some of the workers are Ganapati (1941), Bhuyan (1970), Nasar and Munshi (1974), Khan et al. (1978), Roy (1982), Kanungo (1985), Sarwar and Manzoor (1991), Bose and Gorai (1993), Naik and Purohit (1996), Sinha and Chandravati (1996), Puka and Rao (1997).

During present investigation different water sample of Bankura district were collected of West Bengal state. Present work has been carried out to screen the Bacteria of water bodies of Bankura district and to study the characteristics of micro-organisms causing water pollution with regards to their staining behaviour, colony characteristic and biochemical test.

MATERIALS AND METHODS

Study Site

The Bankura district lies in the western plateau and high lands. Bankura has a tropical climate. In winter, there is much less rainfall in Bankura than in summer. The average annual rainfall of the Bankura District is 1385.9mm.Two sites have been selected for study of micro-organisms and physicochemical characteristics of water. Water sample for study of micro-organisms and physicochemical characteristics were collected from the two sites:

Laboratory media

Dehydrated chemically defined media was used and prepared. Nutrient Agar Media (NAM), Potato Dextrose Agar Media (PDA), MacConkey Agar Media was used to identify the bacterial unknowns in a mixed culture by morphological and biochemical methods. The identification of bacteria is a careful and systematic process that uses many different techniques.

Identification of Micro-organisms

A. Staining (Gram Staining)

Principle

Staining is a simple basic technique that is used to identify micro-organisms. Simple staining is used to study the morphology of all micro-organisms.

B: Biochemical test

- 1. Carbohydrate Fermentation Test
- 2. Urease test
- 3. Indole Acetic Acid (IAA) Production.

RESULTS

Isolation and Identification of Bacterial isolates from Water Samples, Obtained during present investigation were identified on the basis of their Morphological characteristics, Gram's staining and biochemical features.

Isolation of some bacteria and fungi flora

After the continued study from August 09 to April 10, altogether six strains of bacteria were observed and isolate from different Water Samples, Obtained during present investigation. eg:- *E.coli, Staphylococcus, Streptococcus, Psuedomonas, Shigella,* and *Salmonella typhi* Spp. etc. Some fungi species are also isolated from Water samples eg; *Aspergillus* spp.

Isolation and Identification of Bacterial isolates from Water Samples, Obtained during present investigation were identified on the basis of their Morphological characteristics, Gram's staining and biochemical features (Table 1, 2, 3).

Much of ill health which affects humanity, especially in developing countries can be traced to lack of safe and whole some water and food consumption. Since water is vital for our life, we expect it to be clean and safe. The water intended for human consumption must be free of pathogenic and chemical agents, pleasant to taste and usable for domestic purposes. As evident from the selected pond under investigation is highly contaminated with pathogenic coliform and non-coliform bacteria causing pond water pollution which may pose high risk to public health. For ensuring, safe and portable pond water local administration and public health specialist should take some action to preventing water quality problems in pond water with some proper management technique. For examples- Test the pond water periodically to determine bacteria levels and to monitor the presence of any other non-visible problems, strictly limit polluting activities in areas that drain into pond and especially near the pond, use diversion ditches and land grading to divert contaminated surface water away from the pond. The result showed that variation of climatic conditions favours the growth and proliferation

of different physiological types of bacteria. The bacteria numbers were generally higher during the dry season than rainy season. Marshall and Devinny (1988), observed strong seasonal influence on microbial population. The bacteria population being smaller during the cooler, wetter season and the drier months supported large active population. In this study bacterial number were higher during the drier season than the moist seasons. The reason adduced was that during rainy season, lower temperature inhibited bacterial activity, also saturation of the soil by rain limited activity by reducing aeration (Marshall and Devinny 1988). The microorganisms have become a significant attraction as natural source of bioactive molecules with a broad range of biological activities, such as antibiotics, antivirals, antitumorals, antioxidant and anti-inflammatory (Okami, 1982; Nunez et al., 2006; Uzair et al., 2009; Shankar et al., 2010).

S.No.	Form	Colour	Margin	Elevation	Odour
1	Circular	White/ Metallic sheen	Entire	Raised	Faecal odour
2	Irregular Large	Lack of sheen	Entire	Convex	Faecal odour
3	Circular	Golden, Yellow/ White	Entire	Convex	Unpleasant odour
4	Tiny Circular	Grey	Entire	Flat	Unpleasant odour
5	Circular	Fluorescent, Yellowish/ Green Brown Pigment	Undulate	Raised	Fruity odour
6	Circular	Colourless	Entire	Raised	Unpleasant odour

Table 1: Culture characteristics of Bacterial isolates obtained from water sample.

 Table 2: On the basis of all above characteristics bacterial isolates were identified as shown in table.

S. No.	Identified Species			
1	Escherichia coli			
2	Enterobacter aerogenes			
3	Staphylococcus aureus			
4	Streptococcus faecalis			
5	Pseudomonas aeruginosa			
6	Salmonella			

S. No.	No	Name of	Biochemical Test								
2	5. 110.	Sp.	Indole	M.R.	V.P.	Citrate	Urease	TSI	Sugar	Catalase	oxidase
	1.	<i>Shigella</i>	+ve	+ve	-ve	-ve	-ve	A/A no	A/A no	+ve	-ve
	2.	sp. Salmonella sp.	-ve	+ve	-ve	+ve	-ve	gas A/A no gas/ _{H2S} +ve	gas A/A no gas	+ve	-ve

Table 3: Biochemical tests of the isolates.

+= Positive, - = Negative

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