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Microbiological analysis of drinking water quality of Ananthanar channel of Kanyakumari district, Tamil Nadu, India

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ABSTRACT

Bacteriological analyses were carried out on Ananthanar channel water of Kanyakumari district, Tamil Nadu, India. The Ananthanar channel was selected in this study because this channel runs about nearly 28 km and supplies water for many villages for drinking and bathing purposes. Fecal and total coliform counts were performed using the standard membrane filtration technique and multiple tube technique. The results obtained were compared with reports of All India Institute of Medical Sciences Standards for Drinking and Recreational Water. Faecal coliform counts varied from 12 to 180 MPN/100 ml while *Escherichia coli* counts ranged from 6 to 161 MPN/100 ml for all the sampled sites. Among the total coliform *Pseudomonas aeruginosa, Shewanella putrefaciens, Klebsiella pneumoniae, Citrobacter freundii* and *Proteus mirabilis* are reported. The Faecal coliform and the *E. coli* counts exceeding acceptable limits are indicative of pollution from domestic wastes from several informal settlements located along the riverbank. Water uses in the area were determined and were found to be mainly domestic and recreational. The gross pollution of the river exposes the local people who depend on it for their primary water source to serious health risk.

Keywords: *Escherichia coli, Pseudomonas aeruginosa, Shewanella putrefaciens, Klebsiella pneumoniae, Citrobacter freundii, Proteus mirabilis.*

Análise microbiológica da qualidade da água potável do canal Ananthanar do distrito Kanyakumari, Tamil Nadu, Índia

RESUMO

Análises bacteriológicas foram realizadas nas águas do canal de Ananthanar, distrito de Kanyakumari, Tamil Nadu, na Índia. O canal Ananthanar foi selecionado para este estudo porque ele corre por cerca de 28 km e fornece água a muitas aldeias para consumo humano e para fins balneares. Contagens de coliformes totais e fecais foram realizadas utilizando a técnica da membrana de filtração padrão e técnica de tubos múltiplos. Os resultados obtidos foram comparados com os relatórios de *All India Institute of Medical Sciences Standards for Drinking and Recreational Water*. A quantidade de Coliformes fecais variou de 12-180 MPN/100 ml enquanto a contagem de *Escherichia coli* variou 6-161 MPN/100 ml para todos os locais amostrados. Entre todos coliformes *Pseudomonas aeruginosa, Shewanella putrefaciens, Klebsiella pneumoniae, Citrobacter freundii e Proteus mirabilis* estavam presentes. Coliformes fecais e a contagem de *E. coli* que excedem os limites aceitáveis são indicativos de poluição oriunda de resíduos domésticos de vários assentamentos informais localizados ao longo da margem do rio. Os usos da água predominantes na área foram

principalmente domésticos e de lazer. A poluição do rio expõe as populações locais que dependem dele como sua fonte primária de água a um grave risco de saúde.

Palavras-chave: *Escherichia coli, Pseudomonas aeruginosa, Shewanella putrefaciens, Klebsiella pneumoniae, Citrobacter freundii, Proteus mirabilis.*

INTRODUCTION

India is rich in water resources, being endowed with a network of rivers and blessed with snow cover in the Himalayan range that can meet a variety of water requirements of the country (Bhardwaj, 2005). The rivers of India play an important role in the lives of the Indian people. Water resources are great significance for various activities such as drinking, irrigation, aquaculture and power generation. The importance of sustained hydrological studies on Indian waters is now recognized in water resource management due to exploitation of fresh water resources. Report of the scientists at All India Institute of Medical Sciences (AIIMS), New Delhi, finds an alarming prevalence of various diseases causing microbes in drinking water and recreational water. The use of this water may lead to several life threatening diseases. Different authors also reported that Indian River system is polluted mainly because of the human impact (Goel and Bhosale, 2001; Patil et al., 2003; Maity et al., 2004). Significance of water as a potent ecological factor can be appreciated only by studying its physico-chemical and microbial characteristics.

Major factors affecting microbiological quality of surface waters are discharges from sewage works and runoff from informal settlements. Indicator organisms are commonly used to assess the microbiological quality of surface waters and faecal coliforms (FC) are the most commonly used bacterial indicator of faecal pollution (South Africa, 1998). They are found in water that is contaminated with faecal wastes of human and animal origin. Total coliforms (TC) comprise bacterial species of faecal origin as well as other bacterial groups (e.g. bacteria commonly occurring in soil). The coliforms are indicative of the general hygienic quality of the water and potential risk of infectious diseases from water. High FC and TC counts in water are usually manifested in the form of diarrhoea and sometimes by fever and other secondary complications. Bathing and swimming in streams and river are also common among children and adults in the local community. The probability of ingesting infective dose of disease causing microorganism is very high considering the fact that water borne pathogens generally have low infective dose.

Kanyakumari District located in the extreme south of the Indian Peninsula is endowed with substantial number of rivers, rivulets and streams. Ananthanar channel is one among the rivulets of this district. Water from the major river Kodayar is diverted through this channel which takes off from Surlacode Headwork, running for a length of 24 km extended between 77º 39'and 77º 47' of east of longitudes and 8º 07' and 8º 35' north of latitudes. A study on the quality of water of this channel is one of the essential steps as rapid urbanization occurs along the course of this water.

The objective of this work is to evaluate the general bacteriological parameters of the Ananthanar channel of Kanyakumari District, source of water used for drinking and bathing purposes.

2. MATERIAL AND METHODS

2.1. Sample collection

Sample collection is a very important part of river study because conclusions drawn are based only on the testing of collected samples. The purpose of taking samples is to obtain

information, which in some way typifies the aquatic system from which samples are drawn. Grab sampling procedure was adopted as recommended by Standard Method for microbiological analysis. Samples were collected during lean season, on monthly basis, for a period of five months from February 2007 to June 2007. Water samples from three sampling locations of Surlacode, Parvathipuram and Thengampudur and for every sampling date, were collected for this study. Water samples for microbiological examination, were collected in non-reactive borosilicate glass bottles of 500 ml capacity each that had been cleansed and rinsed carefully, given a final rinse with distilled water and sterilized. Samples were taken from the river by holding the bottle near its base in the hand and plunging it, neck downward, below the surface. Then turning the bottle until neck points slightly upward and mouth is directed toward the current. The sampling bottle was not filled up to the brim and 20 mm to 30 mm space was left for effective shaking of the bottle (APHA, 1998). Microbiological analysis of water samples was started as soon as possible after collection to avoid unpredictable changes in the microbial population (Gaudy, 1998).

2.2. Microbiological analysis

Fecal and total coliform counts were performed using the standard membrane filtration technique. The 100 ml water sample was filtered using 0.45 mm pore size, 47 mm diameter filter membrane as described by APHA (1998).

Multiple tube technique was used for the enumeration of Most Probable Number of coliform bacteria. Nutrient agar (NA) as a basal medium MacConkey agar as a differential medium and Blood agar as a special medium were used to determine enteric bacteria. *Escherichia coli* are isolated by inoculating the sample in Bismuth green bile broth. Enteric bacteria isolated on respective selective or differential media were identified on the basis of their colonial, morphological and Biochemical properties (Table 1) following Bergey's Manual of Determinative Bacteriology, 1994.

Table 1. Biochemical analysis.

S. No.	Biochemical Testing	Inferences	Type of Bacteria
1	**Kovacs' reagent:** (Paradimethyl amino bezaldehyde + Isoamyl alcohol + Sulphuric acid)	Appearance of pink coloured ring	Presence of *E.coli*.
2	**Methyl red test:**	Appearance of pink coloured ring in methyl red.	Presence of *E.coli* and *Citrobacter freundii*
3	**Citrase utilization test:** (Simmon's citrate medium + Bromo thymol indicator	Appearance of green colour or blue colour in the medium Green-Negative. Blue- Positive	Absence or presence of *Citrobacter freundii.*
4	**Urease test:** **(**Urease is digested by urease enzyme resulted in release of ammonia)	Appearance of yellow colour shows negative. Appearance of pink colour positive.	Presence of *Citrobacter freundii.* and *Klebsiella pneumoniae*
5	**Oxidase reaction:** (Tetra methyl parapholene diamine dihydro chloride)	Appearance of purple colour within 30 minutes.	Presence of bacteria contains cytochrome oxidase like
6	**Fermentation and gas production test:** (Glucose, lactose, sucrose, mannose	Change of colour from blue to yellow.	Presence of fermenting and gas producing bacteria.
7	**Triple sugar iron test** (Glucose, lactose and sucrose 1:10:10).	Black colour change in the media.	Presence of hydrogen sulphide gas producing bacteria.

Escherichia coli were identified using MacConkey and Brilliant green blue broth as total coliform units in the samples. These two media types support growth of coliforms. *Citrobacter freundii* is capable of using citrate as carbon source for metabolic energy in the absence of fermentable glucose or lactose (Cappuccino and Sherman, 1996; Ashbolt, 2004; Hörman, 2005). Simmon's citrate medium does not have glucose or lactose therefore *E. coli*, which is citrate negative, does not grow on the medium. *Citrobacter freundii* has citrate permease, which facilitates transport of citrate in the cell hence could grow.

3. RESULT AND DISCUSSION

In the present microbial analysis total coliform count and *E. coli* count were analysed for 24 and 48 hours duration. Bacteriological quality of different water samples are shown in Figure 1. In Surlacode (Station I) the Faecal coliform and *E. coli* number is comparatively lesser than the other two stations namely Parvathipuram (Station II) and Thengampudur (Station III) (Figure 1). Different pathogenic coliform were identified in all sampled water (Table 2).

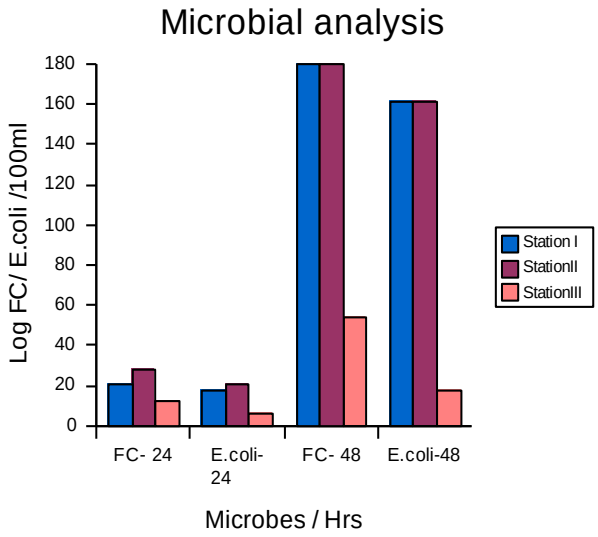

Figure 1. Number of coliform and *E. coli* bacteria in different stations.
Note: FC- Faecal coliform; *E. coli- Escherichia coli*

Table 2. Microbial Isolates from Water Samples

S.No.	Name of the bacteria	Station I	Station II	Station III
1	*Pseudomonas aeruginosa*	+	+	+
2	*Shewanella putrefaciens*	+	+	+
3	*Klebsiella pneumonia*	+	+	+
4	*Citrobacter freundii*	+	+	+
5	*Proteus mirabilis*	+	+	+

The bacteriological analysis of water determines the potability of water. According to Indian standard (BIS, 1981) throughout the year 95% of samples should not contain any coliform organisms or should not be detectable in 100 ml of any two consecutive samples and no sample contains *E. coli* in 100ml. The desirable limit of coliform in water is 10 MPN/100ml (ISI). The result shows that all the water samples in the above three places, i.e. Surlacode (Station I), Parvathipuram (Station II) and Thengampudur (Station III)) were contaminated with high amount of bacterial population than Indian acceptable limit. The reason for high number of bacterial colonies might be due to inadequate maintenance of water reservoirs and the mixing of sewage into the reservoirs or directly into the rivers. In the Station I the high total coliform count may be indicative of the presence of high organic compounds in the water. Because this place is very close to the place of origin and there are so many number of rubber processing industries in this area. In station II and III the primary sources of these bacteria in water are animal and human wastes. These sources of bacterial contamination include surface runoff, pasture, and other land areas where animal wastes are deposited. Additional sources include seepage or discharge from septic tanks, sewage treatment facilities and natural soil /plant bacteria (EPA, 2003). The same results of the high number of total coliforms were observed by different authors in different water bodies in India during pre-monsoon and post monsoon seasons (Rajurkar et al., 2003; Radha Krishnan et al., 2007).

From table 2, it was noted that the water samples had five different types of bacteria. Distinct colonies could be identified using biochemical analysis given in table 1. The results of the bacteriological analysis of drinking water of Ananthanar channel showed that the three areas namely Surlacode, Parvathipuram and Thengampudur, water is contaminated with coliforms and pathogenic bacteria. The bacterial species identified were members of the Enterobacteriaceae family (Table 2). In Surlacode the number of bacterial counts lesser than other areas. One of the reasons may be due to less human intervention. But it is important to note that the limited presence may be due to that coliform bacteria which are widely found in nature and do not necessarily indicate faecal pollution (Binnie et al., 2002; Griffith et al., 2003).

4. CONCLUSIONS

The present study indicates the polluted condition of the water resource which will have serious effects.

Enteric pathogens cannot normally multiply in water hence water is not its mode of transmission to humans (WHO, 1996). However, the presence of enterobacteriae would be enough infective doses in people whose local or general natural defense mechanisms are impaired to significantly low. The people likely to be at risk would be the very old or the very young as well as patients undergoing immunosuppressive therapy. Other immuno-compromised individuals suffering from AIDS would also be at risk. Also, water polluted by bacteria when permitted to contaminate food would lead to the multiplication of the pathogens to very large doses.

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