



ISOLATION AND ENUMERATION OF MICROBIAL INDICATORS IN SHOOLKERE LAKE, BANGALORE.

Environmental Science

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ABSTRACT

Studies are made to evaluate the status of coliform bacteria in the Shoolkere lake, Bangalore, Karnataka. Microorganisms are widely distributed in nature and are found in most natural waters. Water contamination with pathogens and pollutants create many health problems for the people consuming the water in studied area. As such, water quality in relation to human health is an important fact and water borne infections are most common. Considering the increasing urban population and the resulting urban liquid sewage, an attempt has been made to ascertain the bacteriological quality of Shoolkere Lake. The study was carried out for a period of three years (January 2010 to December 2012) giving the monthly variation of Total coliform and Faecal coliform using Membrane Filter (MF) technique. The bacteriological observations indicated the highest peak of total coliform and fecal coliform load showed the maximum count in premonsoon and monsoon season which declined subsequently in postmonsoon season. The bacteriological analysis in the present study indicated that coliform showed significant seasonal variation. The implication of the present result is that the coliform count is above the permissible limits of WHO standards and hence the water poses a health risk to the people consuming it.

KEYWORDS

Total coliform, Faecal coliform, Shoolkere lake.

Introduction

In recent years the pollution of water has become the most significant environmental problems in the world. Water is one of the essential natural resources for existence and development of life on earth. Microbial pollution in natural waters has always been determined by the enumeration of indicator microorganisms. Coliform bacilli and *Escherichia coli* are generally considered to be indicators of the bacteriological quality of water. The surface water sources, in general are not acceptable for consumption as they are often contaminated with different pollutants loaded by organic, inorganic and biological constituents (Kumar *et al.*, 1996; Dahiya and Kaur, 1999). The source of pollutants in water bodies are specifically confined to industrial, domestic or agricultural origin. Industrial pollutants involve the processed water containing heavy metal contaminants, chemicals as well as radioactive compounds (Ibe and Okpleny, 2005). Domestic pollution may involve seepage of septic tank, pit lavatories, cesspools and privies (Omezuruike *et al.*, 2008). Agricultural pollution encompasses irrigational run-offs carrying fertilizers, pesticides, herbicides and faecal matters (Ibe and Okpleny, 2005).

Indicator organisms are commonly used to assess the microbiological quality of surface waters. Faecal coliforms are the most commonly used bacterial indicator for estimating faecal pollution (DWAf, 1996; DWAf, 1998), as they are found in water contaminated with faecal wastes of human and animal origin. Major factors affecting microbiological quality of surface waters are discharges from sewage works and runoff from informal settlements. Faecal contaminated water, if used for domestic or recreational purposes would eventually yield to gastroenteritis, diarrhoea, dysentery, hepatitis, typhoid fever and other fulminant secondary complications (Ballester and Sunyer, 2000). The present study was designed to enumerate the coliform count and to assess the quality of water in Shoolkere lake, Bangalore. Studies on bacteriological aspects of the water bodies have been performed by Thomas *et al.*, (2001), Parihar *et al.*, (2003), Zamkaka *et al.*, (2004), Anderson *et al.*, (2005), Mohan *et al.*, (2007), Krishna H R *et al.*, (2009), and Saleem and Kamili (2011).

Materials and Methods

Samples of water for bacterial analysis were collected at monthly frequency during January 2010 to December 2012. The samples were collected in sterilized borosilicate glass stoppered bottles, the stopper and neck of the bottle should be covered to protect against dust and handling contacts and wrapping paper, pressed over stopper and neck sealed by secure hood. The samples are stored at a temperature between 6–10°C in refrigerator.

Laboratory Analysis

Membrane filter technique was used for counting coliform numbers. This technique involves filtering a known volume of water through a special sterile filter. These filters are made of nitrocellulose acetate and poly carbonate with a 150µm thick and have 0.45µm diameters pores. When the water samples are filtered bacteria in the sample are trapped on the surface of the filter. The filter is then carefully removed placed in a sterile petriplates containing the solidified media and incubated for 20–24 hours at 37°C and 44.5°C for Total coliform and Faecal coliform respectively. The method was followed from APHA (2005).

The enumeration of total coliform and fecal coliform population was made by membrane filter technique with the following high media;

M Endo Media

The medium comprised of the following (g/L): Tryptose 10.0; Thiopeptone, 5.0; casitone, 5.0; yeast extract, 1.5; lactose, 12.5; sodium chloride, 5.0; dipotassium dihydrogen phosphate, 4.37; potassium dihydrogen phosphate, 1.375; sodium lauryl sulfate, 0.05; sodium dosolycholate, 0.10; sodium sulfite, 2.10; basic fuchsin, 1.05; Agar 15.0 and water 1000ml.

M-Fc Media

The medium comprised of the following (g/L): Tryptone 10.0; Proteose peptone No.3 5.0; Yeast extract 3.0; Sodium chloride 5.0; Lactose 12.5; Bile salt No.3 1.5; Aniline blue 0.1g; Agar 15.0 and water 1000ml.

Results and Discussion

Total Coliforms

Bacteria that produce a red colony with metallic (golden) sheen within 24 hours incubation at 37°C on the Endo-type media are considered members of the coliform group (total coliform). The sheen may be cover either the entire colony or may appear only in a central area or on the periphery. The results of monthly variation of Total coliform and Faecal coliform analysis of Shoolkere lake are presented in Fig. 1 and 2. In Shoolkere lake during 2010, the highest total coliforms were observed as (188/100 ml) in April and lowest were observed (43/100 ml) in December. In the year 2011, the highest total coliforms were observed (232/100 ml) in May and lowest were observed (32/100 ml) in October. In 2012, highest total coliforms were observed (366/100 ml) in March and lowest total coliforms were observed (66/100 ml) in September. Total coliform bacteria are a group of easily cultured organisms used to indicate water quality. Total coliform bacteria consist of environmental and fecal types. Coliforms are easy to isolate, present in larger numbers and usually survive longer in an aquatic environment than viruses, parasites and more serious types of bacteria. Most of the total coliforms are not considered pathogens under normal

conditions.

Fecal coliforms

Colonies produced by fecal coliform bacteria on M-FC medium are various shades of blue and nonfecal coliforms colonies are grey to cream-coloured at 44.5°C for 24 hours. In Shoolkere lake during 2010, the highest faecal coliforms were observed (271/100 ml) in August and lowest were observed (118/100 ml) in April. In the year 2011, the highest faecal coliforms were observed (275/100 ml) in October and lowest were observed (131/100 ml) in May. In 2012, highest faecal coliforms were observed (201/100 ml) in June and lowest faecal coliforms were observed (78/100 ml) in March. Fecal coliform bacteria are found in the feces of humans and other warm-blooded animals. These bacteria can enter lakes directly or from agricultural and storm runoff carrying wastes from birds and mammals, and from human sewage discharged into the water. Fecal coliform by themselves are not dangerous (pathogenic). Pathogenic organisms include bacteria, viruses, and parasites that cause diseases and illnesses. Fecal coliform bacteria naturally occur in the human digestive tract, and aid in the digestion of food. In infected individuals, pathogenic organisms are found along with fecal coliform bacteria.

The term "coliform organism" (total coliform) refers to any rod shaped nonspore forming, gram negative bacteria capable to ferment lactose at either 35 or 37°C with the production of acid, gas and aldehyde within 24 – 48 hours, whereas with same properties at a temperature of 44 or 44.5°C are described as "fecal (thermotolerant) coliform" organisms.

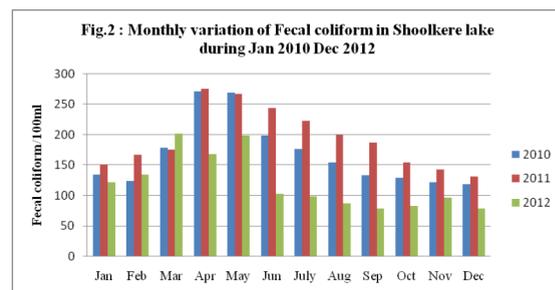
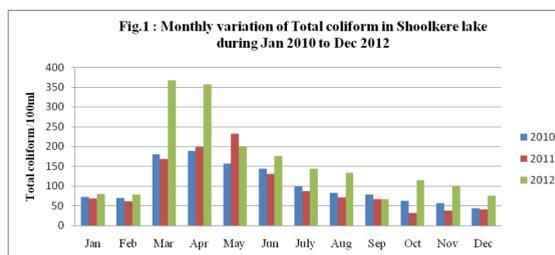
Studies reveal that fecal coliform analysis is a more definitive test for fecal pollution than the total coliforms. The word fecal coliform primarily defines *E.coli* and occasionally *Klebsiella*. It is relatively harmless bacterial species and is almost always present in water that contains enteric pathogens. Thus, because they are relatively easy to isolate and because they normally survive longer than the disease producing organisms, Fecal coliforms are a useful indicator of possible presence of enteric pathogens and viruses. In most cases water that is free from fecal coliforms is considered free of disease producing bacteria. Hence estimation of fecal coliforms is preferred over total coliforms. During the present investigation the total coliform count in the studied lake greatly increased in summer months and decreased in monsoon and winter months, The total coliform count in the Shoolkere lake increased from 2010 to 2012, this increase corresponds to large catchment area fully exposed for the grazing of cattle and other human interferences. Sharma and Mall (1988) and Patralek (1992) opined that temperature also governs the bacterial population. Bacteriological observation for fecal coliform in the studied lake revealed that higher bacterial population with the commencement of monsoon and relatively lower bacterial density during winter. This is in conformity to observations of Parihar *et al.*, (2003) and Mohan *et al.*, (2007). Higher bacterial population during monsoon months was obviously due to transport of organic matter from various sources through surface runoff from the catchment area. This is in accordance with Singh (1985).

With the contamination of water, the aquatic life is also disturbed thus disrupting the whole aquatic system. Enormous anthropogenic activities make all these water resources unfit for consumption. The accumulation of sewage and other waste in Lake, it is not able to recycle them and hence their self-regulatory capability is lost. The decomposition of these wastes by aerobic microbes decreases due to higher level of pollution. Usually the microbiological quality of water is assessed by checking non-pathogenic bacteria of fecal origin. *E.coli* and *Enterococcus Sp.* members are traditionally used as hygienic indicator bacteria (Annie Rompre *et al.*, 2002). Pollution of the water bodies is increasing due to rapid population growth, industrial proliferation, urbanization, increasing living standards and wide ranges of human activities.

Several researchers have pointed out that wastewater treated or untreated is a primary contributor of bacteria to the aquatic ecosystem (Smith, 1970). Water contaminated by bacteria is capable of transferring resistance are of great concern. Since there is the potential for transfer of antibiotic resistance to a pathogenic species (Goyal, *et al.*, 1979). Because of the high incidence of antibiotic resistance among both coliforms and faecal coliforms bacteria they can longer be considered solely as harmless indicators of pollution (Grabow, *et al.*, 1976).

Conclusion

The present study concluded that the contamination in lake water were found generally due to secondary contaminant like discharge of sewage, faecal matter, percolation of industrial waste water etc... Therefore all such water sources should be protected from unhygienic sanitary condition to prevent bacterial contamination and water-borne infections. Hence bacteriology of water continues to be an important component in defining the quality of public health.



REFERENCES

- Anderson, K. L., J. E. Whitlock, and V. J. Harwood. 2005. Persistence and differential survival of fecal indicator bacteria in subtropical waters and sediments. *Appl. Environ. Microbiol.* 71:3041–3048.
- Annie Rompre, Pierre Servais, Julia Baudart, Marie-Rene'e de-Roubin and Patrick Laurent. 2002. Detection and enumeration of coliforms in drinking water: Current methods and emerging approaches, *Journal of Microbiological Methods*, 49, 31- 54.
- APHA. 2005. Standard methods for the examination of water and waste waters, 21st Edn., Washington, DC, USA.
- Ballester, F. and Sunyer, J. 2000. Water and health: precaution must be guided for the health of the public. *J. Epidemiol Community Health.* 54: 729-730.
- Dahiya, S. and Kaur, A. 1999. Assessment of physico – chemical characteristics of underground water in rural areas of tasham subdivisions, Bhiwani district, Haryana. *Environ. J. Poll.* 6(4): 281-288.
- Department of Water Affairs & Forestry (DWAf). 1998. Quality of Domestic Water Supplies-Vol. 1: Assessment guide. Department of Water Affairs and Forestry, Pretoria. pp.4-27.
- Department of Water Affairs and Forestry (DWAf). 1996. South African Water Quality Guidelines. 1: Domestic Water Use (2nd Eds.) Department of Water Affairs and Forestry, Pretoria. pp. 83-87.
- Goyal, S. M., Gerba, C. P. and Melnick, J. L. 1979. Transferable drug resistance in bacteria of coastal canal water and sediment. *Water research.* 13: 349 – 356.
- Grabow, W. O. K., Vanzyl, M. and Prozesky, O. W. 1976. Behavior in conventional sewage purification process of coliform bacteria with transferable or non transferable drug resistance. *Water research.* 10: 717 – 723.
- Ibe, S. N. and Okpleny, J. I. 2005. Bacteriological analysis of borehole water in Uli, Nigeria. *African J. Applied zoology and Environmental Biology.* 7: 116-119.
- Krishna H R, Ramachandra M M and Shivabasavaih., 2009. Microbial quality of total coliforms and fecal coliforms in eutrophicated water bodies of Bangalore Region, Karnataka, India, *The Bioscan Journal*,4(3), 481-486.
- Kumar, A., Bagavathiraj, B. and Bagavathiraj, K. 1996. Physicochemical and Microbiological aspects Choutallam water. *Poll. Res.* 15(2): 159-161.
- Mohan, D., Gaur, A. and Choudhary, D. (2007). Study on Limnology and Microbiology of Naya Talab Jodhpur (Rajasthan). *Proceeding National Symposium on Limnology.* 64 – 68.
- Omezuruike, O. I., Damilola, A. O., Adeola, O. T., Fajobi, Enobong, A. and Olufunke S. 2008. Microbiological and physico-chemical analysis of different water samples used for domestic purposes in Abeokuta and Ojota, Lagos State, Nigeria. *African J. Biotechnology.* 7(5): 617-621.
- Parihar V.L., Sharma M.S. and Sharma L.L. (2003). Utility of Bacteriological Parameters for assessing best use and Trophic status of seasonal waters : A case study from Udaipur, Rajasthan *Poll. Res.* 22(2) : 163 – 167.
- Patralek L.N. (1992). Bacterial density in the Ganges at Bhagalpur, Bihar, *J. Ecobiol.* 3(2): 102 – 105.
- Saleem S., Kamili A.N., (2011) Isolation, identification and seasonal distribution of bacter in Dal Lake, Kashmir, *Inter. j. Envir. Sci.*, 2(1), 185.
- Sharma Arun and Mall Sudha (1988). Bacterial population in three aquatic system of Ujjain. *J. of Hydrobiol* 16 – 19. *Journal of Hydrobiology.*
- Singh, A.K. (1985). Physico-Chemical and Bacterial study of sewage water discharged into the river Ganga at Bhagalpur. *Indian Environ. Ecol.* 3(2): 138 – 142.
- Smith, H. N. 1970. Incidence in river water of *Escherichia coli* containing R – factors. *Nature* 228 : 286 – 1288.
- Thomas Sabu, Harikrishnan k., George Sanil, Paulmurugan R and Das M.R. (2001). Studies on the water quality of Kuttand Wetland ecosystem of Kerala *Poll. Res.* 29 (1) : 59 – 68.
- Zamxaka M, Pironchieva G, Myyiman Y.O. 2004. Microbiological and Physico-chemical assessment of the quality of domestic water sources in selected rural communities of the Eastern Cape Provina, South Africa, *Water SA*, 30, 333.